

In the claims

1. (original) Isolated DNA coding for the CstMI restriction enzyme, wherein the isolated DNA is obtainable from *Corynebacterium striatum*.
2. (original) A recombinant DNA vector comprising a vector into which a DNA segment coding for the CstMI endonuclease has been inserted.
3. (currently amended) Isolated DNA coding for the CstMI endonuclease/methyltransferase, wherein the isolated DNA is obtainable from ATCC Accession No. PTA-5291.
4. (original) A Vector which comprises the isolated DNA of claim 3.
5. (original) A host cell transformed by the vector of claim 2 or 4.
6. (original) A method of producing an CstMI restriction endonuclease and CstMI methylase comprising culturing the host cell of claim 5 under conditions suitable for expression of said endonuclease.
7. (withdrawn) A substantially pure type II restriction endonuclease obtainable from *Corynebacterium striatum* recognizing the following base sequence in double-stranded deoxyribonucleic acid molecules:

5'-AAGGAGN20Ø-3'
3'-TTCCTCN18≠-5'

and having a cleavage position defined by the arrows.

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8. (withdrawn) A method for obtaining Type II restriction endonuclease of claim 7, comprising cultivating a sample of *Corynebacterium striatum* under conditions favoring the production of said endonuclease and separating said endonuclease therefrom.

9. (withdrawn) The Type II restriction endonuclease of claim 7, wherein the restriction endonuclease is purified from GenBank Accession #AAG03371.

IN THE SPECIFICATION

Please replace the paragraph beginning on line 7, page 1 of the specification with the following paragraph:

The present invention relates to a novel type II restriction endonuclease, CstMI. CstMI consists of one polypeptide which possesses two related enzymatic functions. CstMI is an endonuclease that recognizes the DNA sequence 5'-AAGGAG-3' and cleaves the phosphodiester bond between the 20th and 21st residues 3' to this recognition sequence on this DNA strand, and between the 18th and 19th residues 5' to the recognition sequence on the complement strand 5'-CTCCTT-3' to produce a 2 base 3' extension (hereinafter referred to as the CstMI restriction endonuclease). CstMI has a second enzymatic activity that recognizes the same DNA sequence, 5'-AAGGAG-3', but modifies this sequence by the addition of a methyl group to prevent cleavage by the CstMI endonuclease. The present invention also relates to the DNA fragment encoding the CstMI enzyme, a vector containing this DNA fragment, a transformed host containing this DNA fragment, and a process for producing CstMI restriction endonuclease from such a transformed host. CstMI was identified as a potential endonuclease because of its amino acid sequence similarity to MmeI (see U.S. Application Serial Publication No. US-2004-0091911-A1, filed concurrently herewith).

Please replace the paragraph beginning on line 31, page 9 of the specification with the following paragraph:

The MmeI endonuclease was cloned New England Biolabs, Inc. (Beverly, MA) and its amino acid sequence was determined (U.S. Application Serial ~~Publication~~ No. US-2004-0091911-A1, filed concurrently herewith, the disclosure of which is herein incorporated by reference). A BLAST search of the Genbank database using the MmeI endonuclease amino acid sequence as the query returned a number of sequences that were highly significantly similar to MmeI. Among these was a sequence, GenBank accession #AAG03371, which encoded a gene labeled gcrY, and annotated as a "hypothetical 107.5 kDa protein". This hypothetical protein was encoded on a 51,409 base pair plasmid isolated from *Corynebacterium striatum* M82B (see Tauch,A., Krieff,S., Kalinowski,J. and Puhler,A., "The 51,409-bp R-plasmid pTP10 from the multiresistant clinical isolate *Corynebacterium striatum* M82B is composed of DNA segments initially identified in soil bacteria and in plant, animal, and human pathogens" Mol. Gen. Genet. 263 (1), 1-11 (2000)). A sample of this plasmid DNA was kindly provided by the author, Andreas Tauch. The DNA sequence encoding and flanking the potential endonuclease gene was known. Primers were designed to specifically amplify the gene from *Corynebacterium striatum* M82B DNA, with convenient restriction enzyme sites added to facilitate cloning into a vector. The amplified gene was inserted into an expression vector and cloned into an *E. coli* host. Transformed host cells were tested and several were found to express an endonuclease

activity when incubated in NEBuffer 4 supplemented with 100mM SAM (S-adenosyl-methionine) (Figure 7). The DNA recognition sequence of this new endonuclease was determined by mapping the positions of cleavage in pUC19, pBR322 and PhiX174 DNAs. These locations of cleavage were found to be consistent with the sequence 5'-AAGGAG-3' (or 5'-CTCCTT-3' on the complement DNA strand). This novel enzyme was named CstMI (from *Corynebacterium striatum* M82B). This recognition sequence is quite different from that of MmeI, which recognizes 5'-TCC(Pu)AC-3', even though the enzymes share approximately 40% identical and 51% similar amino acids in their sequences (Figure 8). The point of DNA cleavage relative to the recognition sequence was determined by cutting an appropriate DNA with CstMI, purifying the DNA and subjecting it to standard dideoxy automated sequencing. CstMI was found to cleave DNA at the same position relative to its recognition sequence as MmeI; namely after the 20th nucleotide 3' to the 5'-AAGGAG-3' recognition sequence strand, and before the 18th nucleotide 5' to the 5'-CTCCTT-3' recognition sequence strand, producing a 2 base pair 3' extension. CstMI was also found to in vivo modify the recombinant expression vector, pTBCstMI.3, such that it was protected against CstMI endonuclease activity in vitro.

Please replace the paragraph beginning on line 17, page 11 of the specification with the following paragraph:

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In Example II we obtained CstMI by culturing a transformed host carrying the CstMI gene, such as E. coli ER2683 carrying pTBCstMI.3 and recovering the endonuclease from the cells. A sample of E. coli ER2683 carrying pTBCstMI.3 (NEB#1530) has been deposited under the terms and conditions of the Budapest Treaty with the American Type Culture Collection (ATCC) on June 25, 2003 and bears the ATCC Accession No. PTA-5291.